Section II (Remarks)

The claims have been amended to overcome all 35 USC 112 grounds of rejection.

No new matter (35 USC 132) has been added.

Rejection of Claims and Traversal Thereof

In the November 2, 2005 Office Action, claims 1, 3, 4, 6-15 and 18 as previously pending were rejected on reference grounds, including:

- a rejection of claim 7 under 35 U.S.C. §112, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention;
- a rejection of claims 1, 3, 4, and 6 are rejected under 35 U.S.C. 102(b) as being anticipated by Kitagawa, et al. (*Escherichia coli Small Heat Shock Proteins, IbpA and IbpB, Protect Enzymes from Inactivation by Heat and Oxidants*, Eur. J. Biochem., (2002), 269, p. 2907-2917);
- a rejection of Claim 15 under 35 U.S.C. 103(a) as being unpatentable over Lubman, et al. (U.S. Publication No. 2002/0098595 A1);
- a rejection of Claims 7 and 12-14 under 35 U.S.C. 103(a) as being unpatentable over Willsie, et al. in view of Kitagawa, et al. (*Escherichia coli Small Heat Shock Proteins, IbpA and IbpB, Protect Enzymes from Inactivation by Heat and Oxidants*, Eur. J. Biochem. (2002), 269, p. 2907-2917);
- a rejection of Claims 1, 3, 4, 6-11 and 18 under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

These rejections are traversed and reconsideration of the claims is requested in light of the amendments made herein and the following remarks.

Patentable Distinction of Claims as Amended

As a background to the following discussion, it is pointed out that small heat shock proteins are used in accordance with the invention to prevent degradation of proteins in a sample to be analyzed by two-dimensional gel electrophoresis, in which such sample contains protein that is susceptible to degradation by protease, and protease is present in the sample. The instant application specifically defines protein degradation in paragraph [0024] thereof:

"[0024] In the present invention, protein degradation represents a chemical reaction in which a mixture of amino acids or peptides is produced by hydrolysis of peptide bond with protease in vivo or in vitro."

As a result of such degradation of protein by protease in conventional use of two-dimensional gel electrophoresis, the accuracy of the electrophoresis operation for the protein is diminished, and gel spots that would otherwise indicate the presence of the target protein in the sample are reduced, because the target protein is broken up by protein-mediated hydrolysis of the peptide bonds in the target protein.

Although heat shock proteins are known to prevent protein denaturating, denaturing is an entirely different and non-analogous phenomenon – specifically, denaturing is the unfolding of proteins so that conformation is lost. See the present application in paragraph [0012]:

"[0012] Such sHSPs have a conserved region in an evolutionary process and thus has been performing substantially similar functions. ATP independent sHSPs perform a function of preventing protein aggregation irreversibly by combining with denatured proteins under heat stress condition. Therefore, ATP independent sHSPs return the denatured proteins to the original form by correct refolding in cooperation with ATP dependent HSPs(heat shock proteins). For example, it has been reported that IbpA derived from E. coli and IbpB derived from E. coli prevent a citrate synthase from being inactivated by blocking aggregation due to heat or oxidant (Kitagawa et al., Eur. J. Biochem., 269:2907-17, 2002)."

Thus, it has been known that small heat shock proteins block denatured protein aggregation due to elevated temperature (heat) or oxidant conditions, but it has NOT been known prior to the present invention that small heat shock proteins can block protease attack on peptide bonds and

that two-dimensional gel electrophoresis – which does not involve such elevated temperature and oxidant conditions – can be greatly improved in accuracy by incorporating small heat shock proteins in two-dimensional gel electrophoresis samples containing protein that is susceptible to degradation by protease, and where protease is present in the sample.

Kitagawa therefore discloses adding a small heat shock protein to a target protein to block aggregation of the target protein due to heat or oxidant, but does not teach or suggest that such small heat shock protein has additional use for blocking protease attack on peptide bonds in two-dimensional gel electrophoresis samples containing protein that is susceptible to degradation by protease, and where protease is present in the sample.

Accordingly, Kitagawa does not teach or suggest a "gel electrophoresis sample composition that is resistant to protein degradation, comprising protein that is susceptible to degradation by protease, with protease being present in the sample composition, and with the sample composition containing small heat shock protein (sHSP)," as recited in claim 1, or as required in claims 3, 4 and 6.

Lubman discloses that there has been an attempt to understand the proteome (i.e., the quantitative protein expression pattern of a genome under defined conditions) of various cells, tissues, and species and that proteome research seeks to identify targets for drug discovery and development and provide information for diagnostics (e.g., tumor marker).
It also discloses that 2-D gel has been widely used in identifying proteins, including heat shock proteins. However, the mere fact that heat shock proteins can be identified by 2-D gel as the target protein does not in any way teach or suggest heat shock proteins can be added for the analysis of proteomes by 2-D gel electrophoresis. There are numerous kinds of proteins that can be identified by 2-D gel and there is no evidence that one of the skilled in the art would have been motivated to use it for the analysis of proteomes by 2-D gel electrophoresis.

Further, Lubman teaches away the use of 2-D gel in proteome study by expressing that, comparing to 2-D PAGE techniques, his invention provides systems and methods for protein separation and mapping that are highly efficient, amenable to automation, and provide detailed resolution. In *United States v. Adams,* 383 U.S. 39, 52, 148 U.S.P.Q. 479, 484 (1966), the Court

held that "known disadvantage in old devices which would naturally discourage the search for new invention may be taken into account for determining obviousness."

Therefore, claim 15 of the present invention patenably distinguishes over Lubman. Accordingly, withdrawal of this rejection under 35 U.S.C. §103(a) is requested.

Willsie describes the roles of p26 and HSP70 in stabilizing nuclear matrix proteins. It proposed that the majority of p26 translocated into nuclei in response to heat shock, anoxia, low pH and diapause associated with the nuclear matrix, and more specifically, lamin filaments within the nuclear matrix. It also proposed that the association of HSP70 with reassembled lamin filament complexes and p26 indicates that multiple isoforms of p26 and HSP70 may work together in protecting and/or chaperoning nuclear matrix proteins in the embryos. Willsie does not in any way disclose, teach or suggest that p26 can be used to prevent protein degradation.

Further, in Willsie, gel electrophoresis was used as an analytical instrument to examine the association of p26 and HSP70 with nuclear lamins, see page 610, right column, and page 607, right column. Willsie does not in anyway disclose, teach or suggest that p26 can be added to the protein mixture, so as to prevent protein degradation in the 2-D gel electrophoresis.

In Willsie, samples were analyzed by SDS-PAGE for examining the presence of HSP70, HSP90, p26 and lamin, and for examining the association of lamins with HSP70, HSP90, and p26, under control and heat shock conditions, as shown in Figures 2 and 3. Willsie did not suggest any protein degradation process related to small heat shock proteins. Willsie did no experiment involving p26 as inhibitor of protein degradation. Willsie does not in any way disclose, teach or suggest that p26 can be added to the protein mixture to inhibit of target protein degradation.

The deficiencies of Kitagawa have already been discussed.

Accordingly, the reference rejections applied on 35 USC 102 and 103 grounds fail to provide any basis for continued rejection of the claims as now amended.

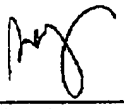
It therefore is respectfully requested that all reference-based rejections be withdrawn.

CONCLUSION

Based on the foregoing, all of applicants' now-pending amended claims are patently distinguished over the art, and in form and condition for allowance. Favorable action is requested.

If any issues remain outstanding, incident to the allowance of the application, the examiner is requested to contact the undersigned attorney at (919) 419-9350 to discuss their resolution, in order that this application may be passed to issue at an early date.

Respectfully submitted,



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